Establishment of Transgenic Creeping Bentgrass (*Agrostis stolonifera* L.) in Non-Agronomic Habitats

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Problems:

- Develop protocols for the Office of Pesticide Programs to assess ecological risks of transgene escape from genetically modified crops into wild plant populations
- There has been little empirical documentation of establishment and distribution of transgenic plants in wild populations.

Approach:

- Survey for *Agrostis* species populations outside the Bentgrass Control Area established by the Oregon Department of Agriculture for test production of GM *A. stolonifera*
- Bulk test 20,400 tissue samples with TraitChek for *CP4 EPSPS* transgenic protein in the field
- Test, locate & sample individual plants expressing *CP4 EPSPS* protein in the field
- Confirm presence of transgene by PCR and DNA sequencing
- Determine species-level parentage of wild transgenics by analyses of biparentally inherited ITS and maternally inherited *matK* DNA sequences

Results:

- Nine transgenic plants were found downwind and up to 3.8 km beyond the control area perimeter
- Transgenic plants were found within wild populations of the same species as the GM crop and within populations of another species that could hybridize with the GM crop
- The parent species of all transgenic plants were the same as the GM crop
- Wild transgenic plants became established at multi-km distance from the GM crop fields due to movement of both transgenic pollen and GM crop seeds

Anticipated Products/Outcomes:

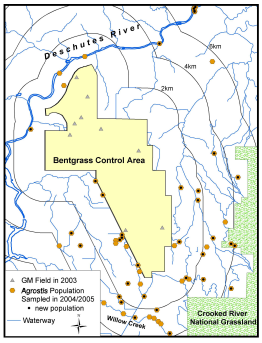
- Experimental protocols for the Office of Pesticide Programs (and other agencies) for post-field release testing/monitoring systems to help inform regulatory decisions regarding the environmental safety of GM plants
- Re-evaluation of isolation distances required between GM crops and compatible wild plant populations

References:

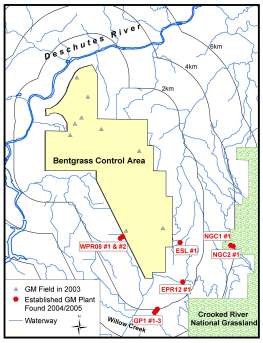
- Reichman et al. 2006. Establishment of Transgenic Creeping Bentgrass (*Agrostis stolonifera* L.) in Non-Agronomic Habitats. Molecular Ecology (in press)
- Watrud et al. 2004. Evidence for landscape-level, pollen-mediated gene flow from genetically modified creeping bentgrass with *CP4 EPSPS* as a marker. PNAS 101: 14533-14538

Abstract

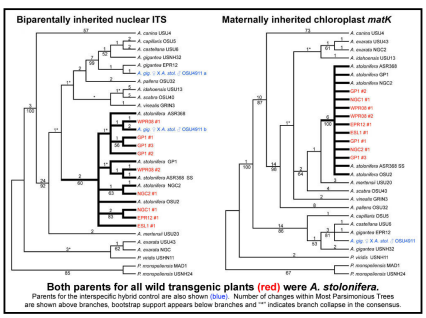
Concerns about genetically modified crops include transgene flow to compatible wild species and potential unintended ecological consequences associated with transgene introgression. To date, there has been little empirical documentation of the relative frequency of establishment and distribution of transgenic plants in wild populations beyond borders of agricultural fields. **We present the first evidence for escape of transgenes into wild plant populations within the USA**; glyphosate herbicide–resistant *Agrostis stolonifera* L. plants expressing *CP4 EPSPS* transgenes were found in Jefferson County, Oregon. Resident populations of three compatible *Agrostis* species were sampled in publicly accessible areas up to 4.8 km outside the control area designated for the test production of glyphosate-resistant *A. stolonifera* (RoundUp Ready creeping bentgrass ASR368, Scotts Seed Company). *CP4 EPSPS* protein and the corresponding transgene were found in nine *A. stolonifera* plants screened from 20,400 samples (0.04%). *CP4 EPSPS*–positive plants were located predominantly in mesic habitats downwind and up to 3.8 km beyond the control district perimeter; two plants were found within the US Department of Agriculture (USDA) Crooked River National Grassland. The spatial distribution and parentage of transgenic plants, confirmed by analyses of nuclear ribosomal internal transcribed spacer (ITS) and chloroplast transfer RNA K intron maturase (*matK*) DNA sequences, suggest that positive plants resulted from both pollen-mediated intraspecific hybridizations and feral plants established from dispersal of crop seeds. These results demonstrate that transgene flow from even short-term field testing can result in establishment of transgenic plants within wild populations. Selective pressure from direct application or drift of glyphosate in mesic habitats where *CP4 EPSPS*–positive *Agrostis* plants are established could enhance transgene introgression and additional establishment. Even without this selective pressure, obligatory outcrossing, vegetative spread by stolons, and dispersal of seed by water, wind, wildlife, or mechanical means could further contribute to the persistence of *CP4 EPSPS* transgenes in wild *Agrostis* populations.



Field Sampling Sites



Location of Wild Transgenic Plants



Parentage of Wild Transgenic Plants

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